

VERSION WITH MARKINGS TO SHOW CHANGES MADE

In the Specification:

The paragraph beginning at page 11, line 25 has been amended as follows:

~~The invention will now be described in more detail with reference to the accompany drawings.~~

Paragraph beginning at page 4, line 21, has been amended as follows:

Figure 1 illustrates the plasmid pHD389; the ribosomal binding sequence, the sequence for the signal peptide from **ompA** and recognition sequence for several restriction enzymes are shown (SEQ ID NO: 14);

Paragraph beginning at page 4, line 24, has been amended as follows:

Figure 2 illustrates the amino acid (SEQ ID NO:3) and nucleic acid sequence (SEQ ID NO:4) for protein LG.

Paragraph beginning at page 5, line 6, has been amended as follows:

Figure 7 illustrates the amino acid (SEQ ID NO:6) and nucleic acid sequence (SEQ ID NO: 5) for protein M1.

Paragraph beginning at page 12, line 5, has been amended as follows:

It has been found that a protein L peptide (expressed in *E. coli*) constructed of the sequence ala-val-glu-asn (SEQ ID NO:15) domain B1 (from protein L) binds to the light chains of the immunoglobulins (W. Kastern, U. Sjöbring and L. Björck. 1992. Structure of peptostreptococcal protein L and identification of a repeated immunoglobulin light chain-binding domain. J. Biol. Chem. 267 (18):12820-5). Since this simple protein L-domain has a relatively

low affinity to Ig, ($1 \times 10^7 \text{ M}^{-1}$), and since the naturally occurring protein L which is constructed of several mutually similar domains (B1-B5) has a high affinity to Ig ($1 \times 10^{10} \text{ M}^{-1}$) four of these domains have been expressed together in the following way:

Paragraph beginning at page 12, line 13, has been amended as follows:

PL-N and PL-C1 are synthetic oligonucleotides (manufactured by the Biomolecular Unit at Lund University (Sweden) in accordance with applicant's instructions) which have been used to amplify a clonable gene fragment which is amplified with PCR (Polymerase Chain Reaction) and which codes for four Ig-binding protein L domains (ala-val-glu-asn-B1-B2-B3-B4-lys-lys-val-asp-glu-lys-pro-glu-glu, SEQ ID NO:1). Amino acids in the protein L-sequence are given for the primer which corresponds to the coded strand (PL-N):

PL-N: 5' -GCTCAGGCAGCGCCGGTAGAAAATAAGAAGAACACCAGAAC-3'

(SEQ ID NO:7)

valgluasnlysglugluthrproglu

(SEQ ID NO:8)

5'-end of this oligonucleotide is homologous with the coded strand in the protein L-gene (emphasized): those codons which code for the last three amino acids in the A-domain (val-glu-asn) are followed by the codons for the first six amino acids in the first of the Ig-binding domains in protein L (B1).

PL-C1: 5' -CAGCAGCA GGATTC TTATTATTCTTCTGGTTTTCGTCAACTT

CTT-3' (SEQ ID NO:9)

Paragraph beginning at page 18, line 13, has been amended as follows:

PL-N and PL-C2 are synthetic oligonucleotides (manufactured at the Biomolecular Unit at Lund University (Sweden) in accordance with applicant's instructions)

which were used, with the aid of PCR (Polymerase Chain Reaction) to amplify a clonable gene fragment, called B1-4, which codes for four Ig-binding protein L domains (ala-val-glu-asn-B1-B2-B3-B4-lys-lys-val-asp-glu-lys-pro-glu-glu, SEQ ID NO:1):

PL-N: 5' -GCTCAGGC~~GGCGCCGGT~~AGAAAATAAGAAGAACACCAGAAAC-3'

(SEQ ID NO:7)

valgluasnlysglugluthrproglu

(SEQ ID NO:8)

P1-C2: 5' -CAGCAGCAGCCATGGTTCTTCTGGTTTCGTCAACTTCTTA-3',

(SEQ ID NO:10)

Paragraph beginning at page 19, line 10, has been amended as follows:

It is known that a simple C-domain from protein G will bind to IgG (B. Guss, M. Eliasson, A. Olsson, M. Uhlen, A.-K. Frej, H. Jörnvall, I. Flock and M. Lindberg. 1986. Structure of the IgG-binding regions of streptococcal protein G. EMBO. J. 5: 1567-1575). The strength at which a simple C-domain binds to IgG is relatively low ($5 \times 10^7 \text{ M}^{-1}$). A fragment which consists of two C-domains with an intermediate D-region having a length of 15 amino acids, however, has a considerably higher affinity to IgG ($1 \times 10^9 \text{ M}^{-1}$). CDC-N and CDC-C are oligonucleotides which have been used as PCR-primers to amplify a clonable DNA-fragment, designated CDC, which codes for two IgG-binding protein G-domains (pro-met-asp-CDC-met).

CDC-N: GG ~~CCATGG~~ ACAC~~T~~ACAAATTAAATCCTTAATGGT

(SEQ ID NO:11)

metaspthrtyrlsleuileleua~~sngly~~

(SEQ ID NO:12)

CDC-C: C ~~AGGT~~CG ACTTATTACATTCAGTTACCGTAAAGGTCTTAGT (SEQ ID
NO:13)

In the Claims:

Claims 14 and 15 have been amended as follows:

14. (Thrice Amended) A protein having the ability to bind to the light chains of immunoglobulins, selected from the group consisting of:

(a) a protein comprising consisting essentially of the amino acid sequence of SEQ ID NO:1;

(b) a protein comprising consisting essentially of the amino acid sequence of at least one of the domains B1, B2, B3 or B4 of (a) wherein,

why this change? (i)(v) domain B1 is comprised of from amino acid 5 to amino acid 80 of SEQ ID NO:1;

(ii)(vi) domain B2 is comprised of from amino acid 81 to amino acid 152 of SEQ ID NO:1

(iii)(vii) domain B3 is comprised of from amino acid 153 to amino acid 224 of SEQ ID NO:1

(iv)(viii) domain B4 is comprised of from amino acid 225 to amino acid 296 of SEQ ID NO:1; and

(c) a protein comprising consisting essentially of the sequence of multiples or mixtures of the domains of B1, B2, B3 or B4 of (b).

15. (Amended) A hybrid protein comprising consisting essentially of one or more of the B1-B4 domains according to claim 14 which bind to the light chains in immunoglobulins of all classes, and domains which bind to heavy chains of immunoglobulin G.